

CLAIMS

What is claimed is:

1. A method of clearing a solution of disrupted biological material, according to steps comprising:

- 5 (a) providing a first silanized silica matrix, comprising a silica solid phase with a plurality of silane ligands covalently attached thereto, wherein each of the plurality of ligands has a neutral charge in a first solution; and
- 10 (b) combining the first silanized silica matrix with the first solution, comprising a disrupted biological material, a target nucleic acid material, and a chaotropic salt at a concentration sufficient to promote selective adsorption of the disrupted biological material to the matrix, thereby forming a first complex.

15 2. The method of claim 1, wherein the disrupted biological material is a bacterial cell lysate.

3. The method of claim 1, wherein the disrupted biological material is disrupted plant matter.

20 4. The method of claim 1, wherein the chaotropic salt concentration in step (b) is at least about 0.5 M.

5. The method of claim 1, wherein the each ligand in the plurality of silane ligands is of the general formula:



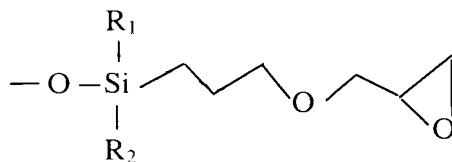
wherein R₁ and R₂ are each a subunit selected from the group consisting of a hydrocarbon chain having from 1 to 5 carbon atoms, an alkoxy having from 1 to 5 carbon atoms, a halogen atom, a hydrogen atom, a hydroxy, an alkyl chain having from 4 to 10 carbon atoms interrupted by an oxy residue wherein up to five of the carbon atoms is substituted by a group selected from the group consisting of a halogen, an alkoxy having

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from 1 to 3 carbon atoms, a cyano having from 1 to 3 carbon atoms, a hydroxy, and a linkage to another silane ligand; and

wherein R_3 is a hydrocarbon chain having from 1 to 20 carbon atoms substituted by at least one hydroxy, an alkyl chain having from 4 to 20 carbon atoms interrupted by at least one oxy group wherein up to ten carbon atoms are replaced by a moiety selected from the group consisting of a halogen, a cyano having from 1 to 3 carbon atoms, an alkoxy having from 1 to 3 carbon atoms, a hydroxy, and an epoxy

6. ~~The method of claim 1, wherein each ligand in the plurality of silane ligands is of the general formula:~~



wherein, R_1 and R_2 are each independently $-OH$, $-CH_3$, $-OCH_3$, or $-OCH_2CH_3$.

7. The method of claim 1, wherein the silica solid phase is a first silica magnetic particle.

8. The method of claim 1, further comprising a step of separating the first complex from the first solution, thereby producing a cleared solution.

9. The method of claim 8, further comprising a step of combining the cleared solution with a second silica matrix in a second solution, wherein the target nucleic acid specifically adsorbs to the second silica matrix, thereby forming a second complex.

10. The method of claim 9, wherein the second silica matrix comprises a plurality of second silica magnetic particles.

11. The method of claim 9, wherein the second silica matrix is a plurality of second silanized silica magnetic particles, and the second solution has a pH of up to about 8.0.

12. The method of claim 9, wherein the first silica matrix and the second silica matrix are the same.

13. A method of clearing a solution of disrupted biological material, according to steps
5 comprising:

(a) providing a first silanized silica magnetic particle comprising a silica magnetic particle with a plurality of silane ligands covalently attached thereto;

(b) combining the first silanized silica magnetic particle with a first solution, comprising a disrupted biological material, a target nucleic acid, and a chaotropic salt concentration sufficiently high to promote selective adsorption of the disrupted biological material to the silanized silica magnetic particle, thereby forming a first complex;
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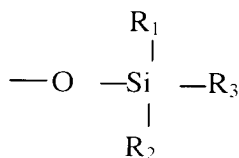
(c) separating the first complex from the first solution, thereby forming a cleared solution.
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14. The method of claim 13 wherein the disrupted biological material is a bacterial cell lysate.

15. The method of claim 13 wherein the disrupted biological material is disrupted plant matter.
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16. The method of claim 13, wherein the first solution further comprises a chaotropic salt at a concentration of up to about 3.5M.

17. The method of claim 13, wherein the each of the plurality of silane ligands is of the general formula:
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wherein R₁ and R₂ are each a subunit selected from the group consisting of a hydrocarbon chain having from 1 to 5 carbon atoms, an alkoxy having from 1 to 5 carbon atoms, a halogen atom, a hydrogen atom, a hydroxy, an alkyl chain having from 4 to 10 carbon

atoms interrupted by an oxy residue wherein up to five of the carbon atoms is substituted by a group selected from the group consisting of a halogen, an alkoxy having from 1 to 3 carbon atoms, a cyano having from 1 to 3 carbon atoms, and a hydroxy; wherein R_3 is a hydrocarbon chain having from 1 to 20 carbon atoms substituted by at least one hydroxy, an alkyl chain having from 4 to 20 carbon atoms interrupted by at least one oxy group wherein up to ten carbon atoms are replaced by a moiety selected from the group consisting of a halogen, a cyano having from 1 to 3 carbon atoms, an alkoxy having from 1 to 3 carbon atoms, a hydroxy, an epoxy, and a linkage to another silane ligand.

10 18. The method of claim 13, wherein the first complex is separated from the first solution in the presence of a magnetic field.

19. The method of claim 13, wherein the first complex is separated from the first solution by centrifugation.

15 20. The method of claim 13, further comprising a step of combining the cleared solution with a second silica matrix in a second solution, wherein the target nucleic acid specifically adsorbs to the second silica matrix, thereby forming a second complex.

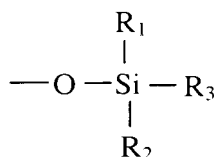
20 21. The method of claim 20, wherein the second silica matrix is a second silanized silica magnetic particle comprising a silica magnetic particle solid phase with a plurality of silane ligands covalently attached thereto.

22. The method of claim 21, wherein the first silanized silica magnetic particle and the
25 second silanized silica magnetic particle are the same.

23. The method of claim 20, wherein the second silica matrix is a silica magnetic particle.

30 24. A method of isolating a target nucleic acid from a nucleic acid adsorption solution, comprising the steps of:

(a) providing a silanized silica matrix comprising a silica solid phase with a plurality of silane ligands covalently attached thereto, wherein each of the plurality of silane ligands is of the general formula:



wherein R₁ and R₂ are each a subunit selected from the group consisting of a hydrocarbon chain having from 1 to 5 carbon atoms, an alkoxy having from 1 to 5 carbon atoms, a halogen atom, a hydrogen atom, a hydroxy, an alkyl chain having from 4 to 10 carbon atoms interrupted by an oxy residue wherein up to five of the carbon atoms is substituted by a group selected from the group consisting of a halogen, an alkoxy having from 1 to 3 carbon atoms, a cyano having from 1 to 3 carbon atoms, and a hydroxy; wherein R₃ is a hydrocarbon chain having from 1 to 20 carbon atoms substituted by at least one hydroxy, an alkyl chain having from 4 to 20 carbon atoms interrupted by at least one oxy group wherein up to ten carbon atoms are replaced by a moiety selected from the group consisting of a halogen, a cyano having from 1 to 3 carbon atoms, an alkoxy having from 1 to 3 carbon atoms, a hydroxyl, an epoxy, and a linkage to another silane ligand;

(b) combining the silanized silica matrix with a nucleic acid adsorption solution having a pH of up to about pH 8.0, the nucleic acid adsorption solution comprising the target nucleic acid and at least one non-target material, wherein the target nucleic acid selectively adsorbs to the silanized silica matrix, thereby forming a complex; and

(c) separating the complex from the nucleic acid adsorption solution.

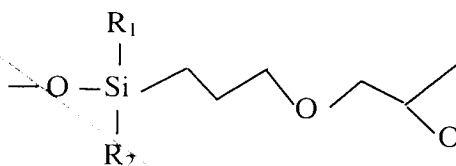
25. The method of claim 24, wherein the nucleic acid adsorption solution comprises a vegetable oil.

26. The method of claim 24, wherein the nucleic acid adsorption solution further comprises a concentration of low molecular weight alcohol sufficient to promote adsorption of the target nucleic acid to the second silanized silica matrix.
- 5 27. The method of claim 24, wherein the adsorption solution further comprises 0.2M to 1.2M of a chaotropic salt.
28. The method of claim 27, wherein the chaotropic salt is selected from the group consisting of guanidine hydrochloride and guanidine thiocyanate.
- 10 29. The method of claim 24, wherein the silica solid phase of the silica matrix is a silica magnetic particle.
30. The method of claim 29, wherein the complex is separated from the nucleic acid
15 adsorption solution in the presence of a magnetic field.
31. The method of claim 24, further comprising washing the complex in a wash solution having a pH of up to about 8.0.
- 20 32. The method of claim 24, wherein the wash solution comprises a concentration of at least about 30% of a low molecular weight alcohol.
33. The method of claim 24, further comprising combining the complex with an elution
25 solution having a pH of at least about 8.0, thereby desorbing the target nucleic acid from the complex.
34. The method of claim 33, wherein the elution solution is a buffer having a pH of at least about 9.0.
- 30 35. The method of claim 24, wherein the target nucleic acid is selected from the group consisting of plasmid DNA, genomic DNA, and total RNA.

36. The method of claim 24, wherein the target nucleic acid is double-stranded linear DNA with a molecular weight of at least about 25 base pairs and up to about 60 kilobase pairs.

37. A method of isolating a target nucleic acid from a nucleic acid adsorption solution using a silanized silica magnetic particle, comprising the steps of:

(a) providing a silanized silica magnetic particle, comprising a silica magnetic particle with a plurality of silane ligands covalently attached thereto, wherein each of the plurality of silane ligands is of a general formula:



wherein, in each formula, R_1 and R_2 are each independently ---OH , ---CH_3 , ---OCH_3 , or $\text{---OCH}_2\text{CH}_3$;

(b) combining the silanized silica magnetic particle with a nucleic acid adsorption solution having a pH of up to about pH 8.0, the nucleic acid adsorption solution comprising the target nucleic acid and at least one non-target material, wherein the target nucleic acid selectively adsorbs to the silanized silica magnetic particle, thereby forming a complex; and

(c) separating the complex from the adsorption solution.

38. The method of claim 37, wherein the adsorption solution has a pH of up to about 8.0.

39. The method of claim 37, wherein the adsorption solution comprises a vegetable oil.

40. The method of claim 37, wherein the adsorption solution comprises the target nucleic acid from an agarose gel slice and the agarose gel.

41. The method of claim 37, wherein the adsorption solution further comprises a concentration of low molecular weight alcohol sufficient to promote adsorption of the target nucleic acid to the silanized silica magnetic particle.

5 42. The method of claim 37, wherein the adsorption solution further comprises a chaotropic salt.

43. The method of claim 42, wherein the chaotropic salt is selected from the group consisting of guanidine hydrochloride and guanidine thiocyanate.

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44. The method of claim 37, further comprising washing the complex in a wash solution having a pH of up to about 8.0.

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45. The method of claim 44, wherein the wash solution comprises a concentration of at least about 30% of a low molecular weight alcohol.

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46. The method of claim 37, further comprising combining the complex with an elution solution having a pH of at least about 8.0, thereby eluting the target nucleic acid from the complex.

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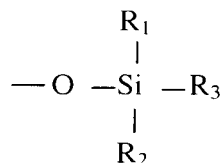
47. The method of claim 37, wherein the target nucleic acid is selected from the group consisting of plasmid DNA, genomic DNA, and total RNA.

48. The method of claim 37, wherein the target nucleic acid is DNA with a molecular weight of at least 25 base pairs and up to about 60 kilobase pairs.

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49. A kit comprising, in a single container:

a plurality of silanized silica magnetic particles comprising a silica solid phase with at least one silane ligand covalently attached to the surface of each particle, the silane ligand having a structure of formula:



5 wherein R_1 and R_2 are each a subunit selected from the group consisting of a hydrocarbon chain having from 1 to 5 carbon atoms, an alkoxy having from 1 to 5 carbon atoms, a halogen atom, a hydrogen atom, a hydroxy, an alkyl chain having from 4 to 10 carbon atoms interrupted by an oxy residue wherein up to five of the carbon atoms is substituted by a group selected from the group consisting of a halogen, an alkoxy having from 1 to 3 carbon atoms, a cyano having from 1 to 3 carbon atoms, and a hydroxy; wherein R_3 is a hydrocarbon chain having from 1 to 10 20 carbon atoms substituted by at least one hydroxy, an alkyl chain having from 4 to 20 carbon atoms interrupted by at least one oxy group wherein up to ten carbon atoms are replaced by a moiety selected from the group consisting of a halogen, a cyano having from 1 to 3 carbon atoms, an alkoxy having from 1 to 3 carbon atoms, a hydroxy, an epoxy, and a linkage to another silane ligand.